Protocol Advisory Subcommittee Report

Protocol to guide the assessment of processing and cryopreservation of male and female gonadal tissue and gametes prior to gonadotoxic treatment to preserve fertility for the future

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Executive Summary

The loss of reproductive function due to cancer or non-malignant diseases, treated with gonadotoxic treatment (chemotherapy, radiotherapy and bone marrow transplantation or surgery to the gonadal tissue or neuroendocrine axis), is a significant survivorship consideration for many patients. The use of gonadotoxic treatment can impact the future fertility of men, women, and children and the late effects consequences of infertility are irrefutable on a patient’s physical and psychological wellbeing.

The sub-specialty of oncofertility has been established to ensure that the reproductive health of all cancer and non-malignant patients receiving gonadotoxic treatment, is considered and if possible preserved prior to starting treatment. Advances in fertility preservation options have allowed fertility to be addressed at earlier stages in cancer care. Increased rates of survival have encouraged clinicians and patients to explore the options available for fertility preservation, allowing the potential for patients to have a biological family in the future with substantial improvements in their satisfaction and quality of life.

This report will discuss all aspects associated with cancer and non-malignant diagnoses which require gonadotoxic treatment that may cause infertility and advancements in fertility preservation options. The report details recommendations focusing on the establishment of three new oncofertility Medicare item numbers:

1. Processing and cryopreservation of ovarian tissue for fertility preservation treatment for female patients.

The FUTuRE Fertility Research Study Group, CanTeen Australia, and our collaborators believe that fertility preservation should be available to all cancer patients and patients with non-malignant disease receiving gonadotoxic chemotherapeutic agents, as a ‘duty of care’ as supported by the Australasian Oncofertility Charter (Appendix 1). The availability of Medicare item numbers will allow equitable access for all Australians of reproductive age, who are diagnosed with a condition requiring gonadotoxic treatment. Appropriate item numbers will ensure that patients have access to consistent oncofertility referral pathways, consultation with a reproductive specialist and the opportunity to undertake fertility preservation, as well as receiving oncofertility follow-up in the survivorship period.
We look forward to hearing about a favorable outcome.

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Chief Investigator FUTuRE Fertility study  
Churchill Fellow 2015
Canteen Australia Summary

For 30 years, CanTeen Australia has supported young people when cancer has turned their world upside down and helped them cope with the physical, emotional and practical impact of living with cancer.

Working with 12-24 year olds, CanTeen supports young people at every stage of their cancer journey, whether they’re dealing with their own cancer or the diagnosis or death of a parent or sibling. Individually tailored support is provided to help every young person deal with the impact that cancer is having on their life, through peer support programs or specialist hospital and community-based services that offer medical care, information and psychosocial support. Monitoring and tracking our programs and services through research and evaluation means CanTeen continually strives to meet the needs of young people affected by the dramatic impact of a cancer diagnosis.

CanTeen is transforming the way young cancer patients are treated through the Youth Cancer Services, which are funded until 2017 by the Federal Government, in partnership with State/Territory health departments. Five Youth Cancer Services across Australia deliver world-class treatment and psychosocial support, ensuring that 15 to 25 year old cancer patients have access to a specialist multidisciplinary team comprising of medical, nursing and allied health support. More than 1,200 young cancer patients were treated and supported during 2014-15. Complementing local service delivery are national strategic priorities in research, data, professional development and advocacy to ensure continuous system improvement and national consistency in models of care, survivorship and other key focus areas.
Summary of the purpose of this document

The applicant has requested the addition of three new MBS item numbers for the discipline of oncofertility in the following populations:

**Population**
1. Male and female patients with any cancer irrespective of stage, who will receive or have received gonadotoxic treatment in three categories: paediatric, adolescent/ young adult (AYA) and adult populations; and
2. Male and female patients with non-malignant disease who will receive or have received gonadotoxic treatment in three categories paediatric, adolescent/ young adult (AYA) and adult populations.

**New Item Numbers**
1. Processing and cryopreservation of ovarian tissue for fertility preservation treatment for female patients.

**Frequency of procedure**
Maximum of one procedure (which may include semen being collected more than once) prior to or after receiving gonadotoxic treatment. Some patients may need to have fertility preservation before and after cancer treatment to ensure an adequate collection.

**Restriction**
Female and male patients who have fertility preservation for nonmedical indication or infertility treatment and who have not received gonadotoxic treatment.
Glossary and Definitions

**Alkylating agents** - activity that inhibits cell division and growth and is used to treat some cancers.

**AMH** - Anti-Müllerian hormone.

**AOFR** - Australasian Oncofertility Registry.

**Andrology** - a branch of medicine concerned with male diseases and especially with those affecting the male reproductive system.

**Anti-Mullerian Hormone (AMH)** - This is a protein released by small pre-antral follicles in the ovary and reflects the follicle pool. Blood tests to check AMH levels may be done as part of fertility testing.

**Assisted Reproductive Technology (ART)** - Methods used to achieve pregnancy by artificial or partially artificial means.

**AYA** – adolescents and young adults, usually aged between 15-25 years old.

**Azoospermia** - absence of sperm in the semen.

**Cancer** - any type of malignant growth or tumor caused by abnormal and uncontrolled cell division.

**Chemoradiation** – chemotherapy followed by radiation to treat cancer.

**Egg** - also known as an ovum, is the female reproductive cell or gamete.

**Embryo** – when an egg and sperm come together (fertilization) they form an embryo, which is the early stage of development of an animal.

**Embryo cryopreservation** – Eggs are collected from a female patient’s ovaries and sperm is inserted into the egg (fertilization). The embryos are then frozen and stored.

**Fertility** - the ability to conceive a baby.

**Fertility preservation** – this is a way to help cancer patients keep their fertility after cancer treatment, in order to have their own biological children.

**Fertilization** – This is the fusion of an egg with a sperm, which leads to the development of an embryo.

**FSH** - Follicle stimulating hormone.

**GnRH analogues** - hormone protection.

**GnRH analogues (GnRHa)** – peptide analogs of gonadotrophin-releasing hormone (GnRH).

**Gonadal organs** - defined as testes or ovaries.

**Gonadal tissue or gonads** - Glands that make sex hormones and reproductive cells; testes in the male, ovaries in the female.

**Gynaecology** - The medical practice dealing with the health of the female reproductive system (uterus, vagina, and ovaries).

**Infertility** - the inability to conceive after 1 year of intercourse without contraception.

**Intracytoplasmic sperm injection (ICSI)** - this is an in vitro fertilization procedure in which a single sperm is injected directly into an egg.

**In Vitro Maturation (IVM)** – This is a method of letting immature ovarian follicles mature in vitro (in a test tube). This method is new and used in a very small number of centres but babies have been born using this method.

**IVF** - In vitro Fertilization techniques.

**MBS** - Medicare Benefits Schedule.

**MSAC** - Medical Services Advisory Committee (MSAC).
**Neuroendocrine axis** - the interaction between the nervous and endocrine systems mainly involving the hypothalamus, pituitary and gonads.

**Obstetrics** - The medical practice of looking after pregnant women during pregnancy and childbirth.

**Oocyte cryopreservation** - egg collection and frozen storage.

**Oncofertility** - Oncofertility bridges the disciplines of oncology and reproductive medicine in order to discover and apply new fertility preservation options for young patients facing fertility-threatening diseases or treatments.

**Ovarian cryopreservation** - the collection and frozen storage of tissue from the ovary.

**Ovarian follicle count** - Ovarian follicles are part of the female reproductive system, and are found in the ovary and decrease through reproductive life to zero at menopause. Each follicle contains a single egg. These eggs are developed only once every menstrual cycle (i.e. once a month in females) until menopause.

**Ovarian tissue cryopreservation** - A whole ovary or tissue from part of the ovary is collected frozen and then stored.

**Ovarian transposition** - surgical movement of the ovaries.

**Ovary** - The ovary is one of a pair of female reproductive organs that produce eggs and release hormones, including estradiol.

**PASC** - Protocol Advisory Sub-Committee.

**POF** - premature ovarian failure.

**Pre-pubertal testicular biopsy** - the collection of immature testicular tissue in pre-pubertal male children, currently experimental.

**Pelvic ultrasound** - This is a type of scan where a probe is rubbed over the lower part of the abdomen (trans-abdominal scan) or inserted into the vagina (trans-vaginal) to look at the ovaries. The probe sends out harmless, high frequency sound waves into the pelvis and an image is formed.

**Psychology** - The study of the mind and of thought, feeling and behaviour.

**Psychologist** - This is a health professional that studies and treats psychological distress.

**Psychological Distress** - This is a term used to describe a range of symptoms and experiences that are commonly held to be troubling, confusing or out of the ordinary.

**Quality of life** - Fertility related well-being.

**Reproductive health** - The health of the reproductive system in its ability to produce gametes (eggs, sperm) and circulating steroid hormones (estradiol, testosterone) to ensure fertility and systemic effects of reproductive hormones.

**Semen** - This is a fluid produced by males that comes out of the penis by ejaculation. The semen contains sperm which can fertilize female eggs.

**Seminoma** - a malignant tumour of the testis.

**Sperm** - The male reproductive cells that combine with female egg cells during fertilization.

**Semen analysis** - To examine semen to measure variables that impact on fertility like semen volume, sperm number, morphology (shape) and viability (motility or swimming speed and directionality).

**Sperm retrieval** - the collection of sperm in post-pubertal men by epididymal or testicular biopsy when semen contains no or too few sperm or sperm cannot be collected by masturbation.

**Sperm cryopreservation/banking** - To collect sperm and then freeze and store for later use.
**Spermatogonia** - a cell produced at an early stage in the formation of spermatozoa.

**Spermatozoa** - male reproductive cells.

**Successful cryopreservation of sperm** - defined as viable sperm recovered after thawing a frozen collection of sperm or semen.

**Trachelectomy** – excision of the uterine cervix.

**Testicular sperm extraction (TESE)** - This is the process of removing a small portion of tissue (biopsy) from the testicle under local anesthesia and extracting the viable sperm present in that tissue.
Background

**Fertility preservation and oncofertility care**

Improvements in the cancer diagnosis and treatment of children, adolescents and young adults, and adult cancer patients of reproductive age (0-44 years) has led to significant improvements in survival rates.[1, 2] As survival rates improve, there is an expectation by clinicians and patients to preserve the reproductive health potential of cancer patients whenever possible.[3-5] A patient’s fertility can be affected by both a cancer diagnosis and cancer treatment (chemotherapy, radiotherapy, bone marrow transplant and surgery).[4, 6-10] which can cause damage to the gonadal organs (testes or ovaries) or the neuroendocrine axis (by inhibiting pituitary hormone secretion that drives gamete production).

A number of studies have shown that infertility following gonadotoxic treatment is a major concern. Potential and actual infertility affects the future quality of life of patients and leads to psychological distress as well as being a predictor of stress in present and future relationships.[3, 11, 12]

Fertility preservation is the overarching term used for medical and surgical treatment to minimise the impact of cancer treatment on a patient’s future fertility by preserving tissue and gametes and protecting fertility during gonadotoxic therapy.[4, 5] There are a number of fertility preservation techniques, which are standard practice and recommended by the 13 international guidance documents on fertility preservation. Currently the fertility preservation options available include:

- Oocyte cryopreservation (egg collection and storage);
- Embryo cryopreservation (fertilization of an egg with either a partner’s or donor sperm);
- Ovarian cryopreservation (the collection and storage of tissue from the ovary – standard of care for adult cancer patients however experimental in children);
- Sperm banking (the collection of sperm or semen via masturbation or testicular biopsy in post-pubertal men);
- Pre-pubertal testicular biopsy (the collection of immature testicular tissue in pre-pubertal male children, currently experimental).
- Gonadal protection during chemotherapy

With the development of fertility preservation strategies and oncofertility care,[13, 14] an increasing number of patients of reproductive age are being referred for fertility preservation and may be able to plan for a biological parentage after cancer treatment.[15]

In 2006, the term “oncofertility” was introduced to describe a new subspecialty focused on the reproductive future for cancer survivors, who may face infertility (as a result of chemotherapy, radiation, or surgery).[16, 17] Oncofertility encompasses: (1) the science needed to develop new fertility preservation options for patients prior to the onset of cancer treatment; (2) the clinical specialties to integrate fertility preservation such as family planning, and hormonal management and (3) advances in oncofertility communication, education and service provisions.[17]
Population at risk of infertility
1. Male and female patients with any cancer irrespective of stage, who will receive or have received gonadotoxic treatment in three categories paediatric, adolescent/young adult and adult populations; and
2. Male and female patients with non-malignant disease who will receive or have received gonadotoxic treatment in three categories paediatric, adolescent/young adult and adult populations.

Gonadotoxic treatments in cancer patients

<table>
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<tr>
<th>Cohort</th>
<th>Age</th>
<th>Number patients diagnosed with cancer annually</th>
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<tr>
<td>Australian population</td>
<td>0-45 years</td>
<td>9,700</td>
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<tr>
<td>Pediatric</td>
<td>0-14 years</td>
<td>600 (6%)</td>
</tr>
<tr>
<td>Adolescent young adult</td>
<td>15-24 years</td>
<td>900 (9%)</td>
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<tr>
<td>Adult</td>
<td>25-45 years</td>
<td>8200 (85%)</td>
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Gonadotoxic treatments in non-cancer patients
Gonadotoxic treatments are sometimes utilised for many non-malignant conditions which pose a major risk of gonadal damage.

Examples of these conditions include:
- **gastrointestinal diseases**[^21]-[^23]. Inflammatory bowel diseases (IBDs), consists of diseases such as ulcerative colitis (incidence rates are 17.4 per 10,000) and Crohn’s disease (incidence rates are 29.3 per 10,000).[^24]
- **rheumatologic disorders** – 6,000 Australian patients of a reproductive age 15-44 years are affected by arthritis and some are treated with gonadotoxic agents.[^25]
- **non-malignant hematologic conditions** – the most common condition treated with gonadotoxic agents is aplastic anemia with incidence rate of 3-5 persons per million in western populations.[^26]
- **autoimmune/vasculitis/glomerular disorders** – 1 per 50,000 Australian patients are affected by vasculitis and 20-150 persons per 100,000 are affected by systemic lupus erythematosus (SLE)[^27]-[^30] most typically diagnosed in females of child-bearing age. Other glomerular disorders including: anti-neutrophilic cytoplasmic antibodies (ANCA) vasculitis and steroid resistant nephrotic syndrome may occasionally require treatment with gonadotoxic agents.
- **gynaecologic conditions** – there are a number of non-malignant surgical conditions eg severe endometriosis, in which surgery may render a patient infertile and fertility preservation at the time of operation will give these women and children an opportunity for a biological family in the future with or without the use of a surrogate gestational carrier.
- **metabolic diseases**[^31]-[^33] – there are a number of metabolic conditions which are treated by the use of bone marrow transplantation and these patients although cured of their metabolic condition are likely to all be infertile.
**Gonadotoxic therapy and male infertility**

In male cancer patients, testicular damage primarily impacts the rapidly proliferating germ cells but at higher doses can also damage and affect the somatic cells of the testis (Sertoli and Lleydig cells)\(^\text{[34]}\). The germinal epithelium of the mature adult testis, where spermatogenesis occurs, is the most rapidly proliferating epithelium in the body, rendering it highly sensitive to gonadotoxic treatments, especially anti-mitotic drugs or irradiation. This germinal epithelium actively produces mature spermatozoa (male reproductive cells), which are susceptible to damage as a result of gonadotoxic treatment.

Cancer treatment at any age can lead to subsequent infertility,\(^\text{[35,36]}\) as the testis can be highly susceptible to damage even before as well as, during, and after puberty. Cytotoxic treatment targets rapidly dividing cells, and as a result spermatogenesis can be disrupted during and after treatment. Recovery of spermatogenesis depends on restoration from germinal stem cells which are also susceptible to cytotoxic damage. The mechanism of this damage is uncertain, but seems to be associated with depletion of the proliferating germ-cell pool, cells at the stage of differentiating spermatogonia.\(^\text{[37]}\)

High dose chemotherapy associated with bone marrow or stem cell transplantation causes severe and often permanent male sterility in most cases.\(^\text{[38]}\) The testis is also highly sensitive to irradiation such that a dose as low as 0.15 Gray causes reduced sperm production and doses of 0.5 Gray or above can cause azoospermia (complete elimination of sperm in the semen). Partial recovery from irradiation-induced azoospermia may occur; however, the time to recovery is proportional to the testicular dose, and may take several years.\(^\text{[39]}\)

**Gonadotoxic therapy and female infertility**

The human ovary has a fixed number of primordial follicles, which is at a maximum at 5 months of gestational age.\(^\text{[7]}\) These follicles are progressively lost with increasing age in an exponential trend, culminating in menopause which on average occurs at around 50 years of age. The rate of oocyte attrition (decline) accelerates rises at around age 35 years. Both chemotherapy, radiotherapy and bone marrow transplantation may lead to an increase in oocyte depletion, leading to premature menopause.\(^\text{[40,41]}\)

Cancer-related infertility, predominantly causing ovarian follicle depletion, is multifactorial and is dependent on a number of factors including, the nature of treatment required but also other intervening variables such as gender, age, pubertal status, gynaecological and reproductive health history, underlying medical conditions, (including genetic or endocrine conditions) and cancer type, and importantly the nature of treatment required.\(^\text{[42,43]}\)

Other factors that may impair fertility or contribute to sub-optimal fertility include damage to the hypothalamic-pituitary-gonadal axis\(^\text{[45,46]}\), immunological and cytological responses to cancer,\(^\text{[47,48]}\) systemic processes (fever, malnutrition and immunosuppression) \(^\text{[37,49,50]}\) and psychological effects of cancer that may affect patient’s sexuality, libido and sexual performance, thereby causing infertility.\(^\text{[51]}\)
Fertility preservation options in males

Options for fertility preservation in post-pubertal males

1. **Semen (masturbation/electroejaculation)**
   
   Cryopreservation (freezing) of semen following masturbation, prior to cancer treatment for male patients, is acceptable and easy to facilitate with minimal or acceptable delay in cancer treatment.[53-74] Currently, cryopreservation of spermatozoa is the most reliable[75] and the only well-endorsed method of fertility preservation in post-pubertal males.[76] This typically involves collecting up to 3 semen samples on alternate days producing up to 50 cryopreserved straws of frozen sperm. Successful cryopreservation of sperm (defined as motile sperm observed after freezing and thawing) is achieved for between 85–100% of male patients; however this is dependent on age (minimum age 14–15+ years depending on pubertal maturity) and diagnosis (including testicular cancer, lymphomas, leukemias, bone cancer and other cancers).[74]

2. **Mature sperm extracted from testicular tissue (open or closed needle testicular biopsy)**
   
   For sperm collection in male patients unable to produce an ejaculate, microsurgical testicular sperm extraction (TESE) is a proven method of sperm collection for cryopreservation.[61, 77-81]

   Future use of sperm can be in the form of intrauterine insemination, IVF-ICSI and possibly testicular tissue grafting. When sperm quantity or quality is limited, intracytoplasmic sperm injection (ICSI) (where an individual sperm cell is injected into an egg cell) is an effective technique to achieve pregnancy.[75]

Options for fertility preservation in pre-pubertal males

1. **Freezing of testicular tissue containing mature and/or immature sperm/spermatogonia**
   
   The pre-pubertal testis does not produce mature spermatozoa. Maturation of spermatogonia from testicular tissue biopsy in prepubescent boys for later clinical use remains in the early phases of experimental research[7, 82, 83] Recent studies have demonstrated that this is feasible in mice and clinical application is likely to be successful in the near future.[84-86] Paediatric fertility preservation should be undertaken under stringent governance at specialised centres.

Fertility preservation options for females

Fertility preservation options for female cancer patients vary depending on the age and gender of the patient, the type and stage of the cancer, urgency of cancer treatment, and whether the patient has a (permanent) partner at the time of diagnosis.[5, 6] Increasing
uptake of fertility preservation options has allowed more patients to preserve their fertility prior to commencing cancer treatment.[52]

**Options for fertility preservation in post-pubertal females**

A number of fertility preservation options are available for post-pubertal women:

1. **Ovarian cryopreservation**
   Ovarian cortical tissue, which contains the majority of the ovarian pool of follicles, can be harvested in an attempt to preserve fertility prior to toxic damage from gonadotoxic treatment.[87]

   Ovarian cryopreservation is utilised in post-pubertal women for multiple indications (1. where there is a high risk of ovarian damage or sterility, 2. prior to conditioning therapy and bone-marrow transplant, 3. when there is not enough time for a cycle of ovarian stimulation, 4. where ovarian stimulation is contraindicated, 5. prior to pelvic surgery, 6. prior to pelvic irradiation).[88]

   In adults, either 1/3 of an ovary or a whole ovary is removed and then processed with slicing into very fine pieces (1 x 3 x 10mm) and cryopreservation. Tissue samples are tested by histology and molecular biological techniques for cancer cells.[89-91] When a woman has completed cancer treatment, and has recovered, but has ovarian failure, the thawed tissue can be transplanted back into the pelvis, to restore ovarian function and normalise levels of gonadotrophins. On average it takes 4-5 months for the graft to begin functioning after surgery. Restoration of ovarian activity was observed in 93% of patients at between 3.5 months and 6.5 months after grafting.[89-92]

   To date, there have been over 100 live births (verbal communication, ISFP 2015 international meeting) worldwide reported after cryopreservation of ovarian tissue in adult patients.[91, 95-110].

2. **Oocyte and embryo cryopreservation**
   Oocyte and embryo cryopreservation are well-established and highly endorsed procedures for fertility preservation in female cancer patients.[76, 111, 112] Ovarian stimulation with storage of the oocytes or embryos that have been created is considered the most reliable fertility preservation technique for post-pubertal women. It requires a woman to undergo stimulation for 8-12 days with regular monitoring and then collection of oocytes under sedation or a general anaesthetic with subsequent cryopreservation of gametes or embryos.[111] There are many reasons to which women may prefer to freeze oocytes instead of embryo including: lack of relationship, relationship uncertainty, maintaining complete autonomy regarding future use of gametes, and potential ethical as well as religious concerns.

   For patients who have a partner, embryos can be created using in vitro fertilization techniques and then frozen.[17, 113, 114] The term cryopreservation refers to the storage of viable cells at low temperatures (normally at −196°C).[115] The ultra-rapid cooling method also known as vitrification[115] has resulted in an increase in the success rate of both oocyte and embryo cryopreservation.
Additional considerations have to be made by patients, families and specialists before patients undergo oocyte stimulation, such as the potential effects of any delay in oncological treatment and decisions about the best stimulation protocols that limit exposure to increased estrogen levels induced by ovarian stimulation.

3. **Gonadotrophin releasing hormone (GnRH analogues)**

GnRH analogues (hormone protection) have been utilised during chemotherapy to suppress ovarian cyclicity and reduce the accelerated recruitment and atresia which occur in response to the chemotherapy. GnRH analogues induce a temporary medical menopause in an attempt to protect the ovaries from the gonadotoxic effects of chemotherapy.[7,116-118]

One class of GnRH analogues (GnRH agonists) induce an initial supraphysiological release of gonadotropins and within several days they desensitize the GnRH receptors on the pituitary gonadotropes, preventing the endogenous pulsatile GnRH to exert its physiological action, resulting in a hypogonadotropic state which aims to replicate the pre-pubertal state of the neuroendocrine axis phenomenon.[118] However, the hypoestrogenic state may induce menopausal symptoms, such as hot flushes, vaginal dryness and sleep disturbance, more common in older and infrequent in younger patients. By contrast, GnRH antagonists directly induce the down regulated hypogonadotrophic state without any initial release.

Over 20 studies (including five prospective randomized controlled trials) have reported on patients treated with GnRHa during chemotherapy, mostly but not universally showing a significant decrease in premature ovarian failure (POF) rate in survivors.[116,119-129] Studies have reported that > 90% of patients treated with GnRHa during chemotherapy maintained ovarian function, with a pregnancy rate of approximately 19%,[123] 22% suggesting that the use of GnRHa co-treatment can help to preserve not only ovarian function but also fertility in the medium term.[123]

4. **Ovarian transposition**

Ovarian transposition (surgical movement of the ovaries) also known as ovarian suspension, oophoropexy, or ovariopexy)[130, 131] may be used for fertility preservation in women receiving pelvic radiation.[130,131] Ovarian transposition is a surgical technique used to protect ovarian function before a patient receives radiation. This procedure aims to move the ovary out of the irradiation field, protecting it from direct radiation and irreversible damage thereby preserving its function. Laparoscopic ovarian transposition in women <40 years of age is associated with preservation of ovarian function in 88.6% of cases.[132] Studies have reported that 90% of patients who had ovarian transposition before radiotherapy resume normal levels of follicle-stimulating hormone and estradiol.[133]

**Current options for fertility preservation in pre-pubertal females**

1. **Ovarian cryopreservation**

The only method of fertility preservation in pre-pubertal female children is ovarian tissue cryopreservation. The same method is used for children as for adults but the procedure is technically more challenging because of the size of the patient’s
ovaries. The main aim of this strategy is to ultimately re-implant cortical ovarian tissue into the pelvis once treatment is completed and the patient is disease-free. For children undergoing this procedure, small slices of (4 x 5 x1 mm) are frozen in individual vials and stored in liquid nitrogen (cryopreserved). The added advantage of re-implanting ovarian tissue in children is induction of puberty. Frozen ovarian tissue not only retains reproductive potential, but also the functional unit of the ovary, the follicle. Follicles in the transplanted tissue possesses the capacity to produce estradiol and other sex hormones that maintain regular menstrual cycles. Sex hormones exert a plethora of different functions in the female body and maintained female steroid producing capacity opens new possibilities.

Internationally a number of centres offer ovarian tissue storage for pre-pubertal girls, within ethical frameworks. To date there has only been one birth from a child who had ovarian tissue stored at 13 years of age prior to bone marrow transplant but numbers are expected to rise imminently. Institutional ethics committee approved protocols are required for paediatric fertility preservation, which should only be undertaken under stringent governance at specialised centres.

Fertility related psychological distress
The loss of reproductive function is one of the most troubling adverse consequences of successful curative cancer treatment, causing psychological distress and diminishing the quality of life of cancer survivors.

Cancer patients have a strong desire to be informed about the available options and strategies associated with fertility preservation but research shows that many patients are not provided with information about fertility preservation options and strategies.

Infertility can have long lasting psychological ramifications on the quality of life for cancer patients, who have not yet started or completed their family at the time of their cancer diagnosis. Cancer survivors whose fertility has been compromised by their treatment, experience heightened psychosocial and emotional distress and have more difficulties in adjusting to life after cancer than those who do not lose their reproductive capacity.

Current barriers for uptake of Fertility Preservation
Despite promising advances in technology in the past decade and an increasing number of patients seeking fertility preservation, several clinician and patient barriers exist in providing fertility preservation:

Available Clinical Information
There is not yet good quality data on the short and long term effects on fertility of for the new novel chemotherapy agents or combinations of chemotherapy or multimodality treatment. A lack of current and relevant information can be a potential barrier for patients receiving the specialty care they require.
Provision of written as well as verbal information about the possible effects on fertility of cancer treatment at the time of a cancer diagnosis is required because at that time a patient's focus is on processing information associated with their cancer diagnosis.[146, 160, 161] However, various studies demonstrate that not being given the opportunity to discuss fertility preservation at any stage throughout the cancer trajectory causes heightened psychological distress.[3, 11, 12]

Available Patient Information
Unfortunately, less than 50% of cancer patients[146, 158, 162-166] report being informed about potential risks to fertility associated with their cancer diagnosis, and less than 35% of cancer survivors recall discussing the possible risks of pregnancy during or after cancer treatment, or available fertility preservation options, with a health care provider.[3, 4]

Some cancer specialists feel uncomfortable broaching the topic of sexual health, particularly if sexual and reproductive health is outside their realm of expertise.[150] Other studies have reported oncologists’ non-referral relates to a deviation from their primary objective, which is to treat the patient’s malignancy.[167] Concerns regarding delaying a patient’s cancer treatment have also been documented as a concern and potential barrier for referral from cancer to reproductive specialist. [168] Health care professionals also report lack of knowledge, skills and training associated with discussing fertility preservation, as well as a lack of standardised guidelines available in Australia for referral.

Conversely, some cancer patients and their families may be focused solely on survival and may not consider the future impact of the cancer treatment on the patient’s fertility. However, current studies report that parents, family members[169, 170] and young cancer patients[3, 146] would like to have discussions regarding their cancer and its implications on their reproductive health.

Health care professionals also report lack of knowledge, skills and training associated with discussing fertility preservation, as well as a lack of standardised guidelines for referral in Australia.[171-173]

Barriers for rural patients and non-English speaking patients
Non-English speaking patients face additional barriers; fertility preservation information is mostly not discussed at the point of consultation by the cancer specialist. [165, 169, 174] Uptake for Assisted Reproductive Technology services are predominately utilised by affluent English speaking patients.[175-177]

Patients who are treated in rural cancer centres have additional barriers in accessing fertility preservation, as fertility and andrology centres are usually based in metropolitan or regional locations.[178-181]

Referral pathways
In Australia there are nationally agreed guidelines for fertility preservation, however we do not have state or national referral pathways and this results in some patients missing out on the opportunity for fertility preservation.[164, 182, 183] Referral pathways between clinics exist but they are often ad hoc between certain clinicians. Unfortunately, many cancer specialists indicate that they are unaware of whom or where to refer a patient for fertility preservation services and this is a barrier for referral.[169, 174, 184] This is especially...
relevant for patients residing in rural and regional areas, where access to fertility preservation services is limited.

**Specialist advice**

Most cancer patients receive information about fertility preservation by cancer experts and not fertility and andrology specialists but unfortunately not all cancer doctors inform patients about the potential of losing their fertility \(^{[148, 185, 186]}\) or do so on an ad hoc basis, even though they recognise the importance of providing this information.

In 2005 the ethics committee of the American Society for Reproductive Medicine extended physicians’ duty to ‘inform patients about options for fertility preservation and future reproduction prior to treatment.’\(^{[76, 170]}\)

Regrettably in some areas, there is still suboptimal communication between cancer and fertility specialists.\(^{[187]}\) Given the competing demands of providing complicated and detailed information about fertility potential and the risk to fertility, based on cancer treatment, there is a role for cancer specialists to work collaboratively with reproductive and andrology specialists to achieve the best outcome for patients.\(^{[60, 188]}\)

**Timing**

Timely referral and uptake of fertility preservation is important and so is early commencement of treatment with gonadotoxic agents. Delays in referral to fertility preservation services and the length of fertility preservation treatment, are often reasons for why patients choose to start treatment prior to referral to fertility preservation experts. In one study only 20% of parents and 30% of adolescent patients indicated they would delay treatment to undergo fertility preservation methods \(^{[148, 189]}\) but it is\(^{[148, 189]}\) unclear to what extent clinicians views influence decisions.

**Costs**

Fertility preservation treatments are expensive; the upfront cost and the lack of Medicare and insurance coverage for some aspects of oncofertility care is often a reason for clinicians’ lack of referrals and for patients not taking up referrals to a fertility or andrology specialist. The cost of processing, cryopreservation and storage are not included in the current Medicare Schedule and are a significant barrier for patients.

The American Medical Association (AMA) adopted a new policy (resolution 6) in 2012 supporting financial cover of fertility preservation when iatrogenic infertility may be caused, directly or indirectly by medical treatment necessitated for cancer therapy.

The guidelines from the National Comprehensive Cancer Network\(^{[190]}\), American Society of Clinical Oncology, the guidelines from the National Comprehensive Cancer Network\(^{[190]}\), American Society of Clinical Oncology\(^{[170]}\) and Clinical Oncological Society of Australasia \(^{[191]}\) agree that potential fertility side effects of treatment must be discussed, and fertility preservation should be offered. Without Medicare and insurance coverage, these guidelines are an unattainable recommendation for many patients.
Handling, cryopreservation and storage of ovarian tissue summary box

Preparation of the cortical ovarian tissue
The ovarian tissue is prepared prior to freezing by dissecting apart the surface (cortical) tissue containing the follicles and the inner part mainly circulation and support tissue. The surface tissue is subsequently dissected into 1mm thick slices to facilitated movement of the cryoprotectants (anti-freeze solutions). This is a manual procedure. This takes several hours.

Freezing of the cortical ovarian tissue
The slices of ovarian cortical tissue are exposed to cryoprotectants to remove water from the cells and placed in vials in an automated freezing machine which gradually reduces temperature at a controlled rate over time to -150 °C. The vials are then stored in a large tank containing liquid nitrogen at a temperature below -150 °C. This takes several hours.

Storage
The vials must be maintained at a temperature of -150 °C to preserve the cellular integrity, and this is achieved by replenishing the liquid nitrogen reservoir in the storage tank.

Ovarian tissue can be obtained during laparoscopy or laparotomy under general anesthetic. Once the tissue is removed, it will be analysed for detection of gametes and also evaluated by histological and molecular biological techniques to exclude malignant cells. If any mature eggs are present they will be removed and frozen in liquid nitrogen. Ovarian tissue can be frozen using three different approaches: most commonly tissue is prepared as fragments of ovarian cortex, since there are still technical challenges (relating to cryoprotectant dissemination in tissue) precluding the widespread practice of freezing of a whole ovary and its vascular pedicle. Isolated follicles can also be cryopreserved. Following ovarian tissue retrieval, the tissue is processed, placed in straw like tubes, which are then placed in a cryopreservant (an antifreeze solution) and stored at subzero temperatures, until they are required for use.

The procedure includes the following:
1. Before collecting tissue remove freezing solutions from fridge and equilibrate to room temp.
2. Place tissue in large petri dish containing Hepes (20 ml). Examine and record information; size, surface; appearance, colour, presence of blood on surface, density, follicles, CL, medulla, appearance, density, abnormalities, follicles.
3. Puncture with fine needle any follicles, expel follicular fluid by applying pressure search for oocyte cumulus mass. Transfer oocytes to Nunc well, containing Fert medium. These will be cultured in maturation medium prior to freezing.
4. Before trimming remove small area of tissue for histology (1/10). Preference; take poorer area i.e. bloody area, area containing corpus luteum, irregular surface, odd looking. Place in labelled container in 10% formalin. For query ovarian cancer or endometriosis take piece from each end and area abutting tissue which will be frozen.
ie middle (place only one piece in each path pot and number same as pieces). Track the piece of origin against slices in vials (record).

Prepubertal Tissue (≤ 15 years)

1. All of the medulla is to be frozen for the patient. Remove medulla in one piece, slice this into roughly 1-2 mm slices treat in similar way to cortex slices.
2. Only prepare cortex in slices, no blocks are to be cut. Record which vials contain cortex (5 slices/vial) and medulla (2 pieces of medulla /vial).
   1 vial is to contain 1 piece of medulla for testing another to contain 2 slices of cortex for testing. Include these in patient total. 1 slice of cortex for fixing.

Adult Tissue

1. Cut away medulla using scissors and forceps (aim 1 mm thick), remove all corpus luteum and any bloody areas. In tissue from leukemia /lymphoma patients remove completely all blood containing areas.
   Retain two large blobs of medulla to be frozen for later assessment of malignant contamination of tissue (freeze in 1 vial).
2. Cut cortex into 4 -5 rectangular pieces (~5mm wide). Working with one piece at a time. Cut into ~1mm slices using double sided razor blade. Use grid under scope to check size. For whole ovary cut some into small blocks 5x5 mm again check size with grid. At completion of each piece, transfer slices to sieve sitting in 8 mls Fert Medium (CO₂ ) in small petri dish.
   Record number of slices in each sieve (~ 15 slices per sieve, 2 blocks per sieve). For smaller amount of tissue (<50 slices) put 10 slices per sieve.
   1-2 slices of cortex to be given to Kelly/Renee for fixing and any small irregular pieces.
3. Check dishes for oocytes, move to Fert medium. These will be cultured in maturation medium prior to freezing.
4. Using 6 well plates label and aliquot freezing solutions (8ml). Left side PBS +alb, right side 1.5M PROH +0.1M Sucrose +alb. Dehydration procedure conducted at room temperature.
5. Remove sieve from medium with forceps touch side of sieve on dish lid to remove excess medium. Dip sieve in PBS well, remove from PBS (<30sec) drain on side of well, transfer sieve to 1.5 MPROH +0.1Sucrose +alb, lift to remove any bubbles trapped under sieve. 2 bloody areas of medulla are also dehydrated in solutions. Periodically swirl sieve and using forceps move slices around in sieve (tend to clump together and move to side of sieve). Leave in 1.5 PROH+0.1 Sucrose +alb for 90 minutes.
6. During this time label vials with UR, Surname, Freeze Date, Vial number and put coloured plugs in lids with number on top. Place 0.5 ml of 1.5 M PROH +0.1 M Sucrose +alb in each vial. See above for number of slices /vial. 1 vial to contain 2-3 slices of cortex for testing if required later (record in patient total).
7. At about 80 min start to load slices into vials. Load vials onto canes start freeze program at 90 min. Use Embryo 1.5 PROH program. Seed at -7 °C using jumbo swabs, check for ice nucleation.
8. Record all information in red book and ovarian tissue sheet.
   Complete letter, make 3 copies; 1 to each of following original to patient, referring doctor, Kate, one to be file with ovarian tissue sheet.
Storage
- Large Vapour Tank. Fill lowest level boxes first.
- Record position, cap colour, number vials.
- Mark off position on rack box sheet.

Freezer Program
- **Planner Freezing Machine**
- Embryo Freezing 1.5 PROH Program
- Pause 16 °C
- Ramp 1: -2 °C /min to -7 °C
- Ramp 2: Hold at -7 °C for 10 min
  manual seed with giant swabs, watch ice formation
- Ramp 3: -0.3 °C /min to -30 °C
- Ramp 4: -50 °C /min to -150 °C
- Ramp 5: Hold at -150 °C for 2 hours

Cryostorage refers to the process of freezing the ovarian tissue with a view to thawing the tissue for use in assisted reproduction treatments when the patient is ready for family planning. There are two methods of cryopreservation: slow freezing and vitrification. Slow freezing refers to the exposure of the tissue to cryoprotectant and cooling the tissue slowly in approximately -140°C, after which time the tissue is put into liquid nitrogen at -196°C for storage. Vitrification of ovarian tissue is a rapid method of cryopreservation developed to eliminate the risk of ice crystal formation in ovarian tissue. Vitrification differs from the slow-freezing method in the concentration of the cryoprotectant (high) and the rate of cooling (fast, within minutes).[192-199]
Handling and storage of testicular tissue

Handling, cryopreservation and storage of male testicular tissue and summary box

Preparation of the testicular tissue
Testicular tissue is collected during a short surgical procedure performed under general anaesthetic. The testicular tissue is then prepared for cryopreservation in the laboratory by an Andrologist where it will be processed and then frozen.

Cryopreservation of testicular tissue and sperm
Tissue pieces are placed in 1.8ml cryovials containing 1.5 ml of cryoprotectant medium, which consisted of 5% DMSO and 5% human serum albumin (HSA) solution diluted in HBSS. Equilibration is then performed at +4°C for 30 min. The samples are then cooled in a programmable CL863 freezer. Testicular tissue freezing can be done by either slow freezing or vitrification (rapid freezing) of immature testicular tissue.

Storage
Samples are placed on a vitrification block or plate precooled to -196°C with liquid nitrogen. The sample will instantly freeze on contact with the surface.

The vitrified samples are then transferred to a liquid nitrogen-cooled cryovial before storage in a liquid nitrogen tank. For DCV, the fragments are immediately put in a cryovial and directly exposed to liquid nitrogen.

Testicular tissue freezing can be done by either slow freezing or vitrification (rapid freezing) of immature testicular tissue.

Testicular tissue collection
Testicular tissue is collected during a short surgical procedure performed under general anaesthetic. During the operation, a urologist collects a wedge-shaped section (biopsy) from one of the testes. The size of the biopsied tissue is usually about 1.2 × 2.7 × 9.10 mm. The testicular tissue is then prepared for cryopreservation in the laboratory by an Andrologist where it will be processed and then frozen.

The Andrology scientist will collect the sample directly from the surgeon as soon the tissue is removed. The biopsy is placed directly into a physiological buffered collection media containing HEPES and 5% human serum albumin (HSA) or equivalent solution in a MEA toxicity tested specimen container. The vessel is then labelled with the patient’s addresso, displaying a minimum of three patient identifiers. The testicular tissue is then transported for cryopreservation in the laboratory where it will be processed and frozen (a chain of custody must be maintained at all times for such samples; from the time of collection in the theatre, during transport, cryopreservation and cryostorage).

Cryopreservation of testicular tissue and sperm
Tissue pieces are placed in 1.8ml cryovials (NUNC, Life Technologies, Roskilde, Denmark) containing 1.5 ml of cryoprotectant medium, which consisted of 5% DMSO (Sigma-Aldrich, Sweden AB) and 5% human serum albumin (HSA) solution (Vitrolife, Goteborg, Sweden)
diluted in HBSS. Equilibration is then performed at +4°C for 30 min. The samples are then cooled in a programmable CL863 freezer.

Testicular tissue freezing can be done by either slow freezing or vitrification (rapid freezing) of immature testicular tissue.\textsuperscript{256}

**Controlled slow freezing**

Using aseptic techniques at all times the tissue is removed from the transport medium and the segment dissected into smaller fragments, increasing the surface area of the tissue to ensure sufficient contact and penetration of the cryoprotectant reagent. A number slow freeze cryopreservatives and methods have been identified.

1. Tissue segments are placed in 1.8ml cryovials (NUNC, Life Technologies, Roskilde, Denmark) containing 1.5 ml of cryoprotectant medium, which consisted of 5% DMSO (Sigma-Aldrich, Sweden AB) containing 5% HSA solution (Vitrolife, Göteborg, Sweden) diluted in HBSS.

2. Tissue pieces in cryovial filled with sterile Hank’s balanced saline solution (HBSS; 14175-129; Life Technologies, Merelbeke, Belgium), containing 0.7 M DMSO (D2650; Sigma-Aldrich, Bornem, Belgium) without sucrose and 5 mg/ml human serum albumin (HSA; 10046; Vitrolife, Göteborg, Sweden) or with 0.1 M sucrose (+S; 10274-5c; VWR, Leuven, Belgium) and 10 mg/ml HSA.

3. Alternatively, the tissue can be loaded into a series of 0.3ml CBS high security cryostraws (Cryo Bio Systems, Groupe IMV Technologies, L'Aigle, France), in 1.5M Propanediol, 0.5M Sucrose, 1% human serum albumin (ART-8014, SAGE media, Trumbull, USA), with no DMSO and heat sealed.

Cryoprotectant equilibration is then performed at +40°C for 30 min.

Using a programmable freezer, vials are cooled at 1°C/min with holding at 0°C for 5 min, followed by cooling at 0.5°C/min until −8°C. At this temperature, the program is put on hold for 10 min to allow manual seeding. The program continues at a rate of 0.5°C/min until −40°C, held for 10 min, and continues to −70°C at 7°C/min, with subsequent plunging to liquid nitrogen.

**Thawing**

The samples are thawed in a water bath at 37°C until the ice melts (2 min) and then washed twice for 5 min in sterile HBSS on ice or, when applicable, in a reversed sucrose gradient solution (0.1, 0.05 and 0 M sucrose).

**Vitrification**

Using aseptic techniques at all times the tissue is removed from the transport medium and the segment of tissue dissected into smaller fragments, increasing the surface area of the tissue to ensure sufficient contact and penetration of the cryoprotectant reagent.

The testicular fragments are exposed to a DMEM/F12-based vitrification solution containing 1.05 M DMSO and 1.35 M ethylene glycol (EG; E-9129; Sigma-Aldrich) for 10 minutes and 2.1 M DMSO and 2.7 M EG for 5 min each. At the second step, the vitrification solution is supplemented with 20% HSA.
Tissue fragments are then removed from the last step media and placed one at a time in direct the vitrification block or plate precooled to -196°C with liquid nitrogen. The tissue fragment will instantly freeze on contact with the surface.

The vitrified tissue samples are then transferred to a liquid nitrogen-cooled cryovial before storage in a liquid nitrogen tank. For DCV, the fragments were immediately put in a cryovial and directly exposed to liquid nitrogen.

**Thawing**
Samples are warmed by adding a pre-warmed solution (37°C, DMEM/F12 + 0.5 M sucrose + 20% HSA) to the cryovials and by keeping the samples at 37°C for 2 min. Samples are then washed in DMEM/F12 with 20% HSA for 2 min at 37°C and finally immersed in a fixative for light or electron microscopy.[257]

**Tissue Histology**
Small fragments of the biopsied tissue must be sent for histopathology to check the nature and quality of the tissue and to ensure that the frozen material is free of any disease. This must be completed before any attempts of propagation and/or autotransplantation of spermatogonial stem cells.
Regulatory Information

Reproductive Technology Accreditation Committee (RTAC) Certification

All fertility and andrology centres are licensed by the Reproductive Technology Accreditation Committee (RTAC) Certification Scheme, developed by the Fertility Society of Australia and independently audited by JAS-ANZ (see Table 1). This is an independent body which is responsible for ensuring certain minimum standards are met by all fertility and andrology clinics in Australia.

RTAC is an independent body responsible for ensuring standards are met by all fertility and andrology clinics in Australia. All processes, from clinics to laboratories and day hospitals, will have ISO 9001:2008 accreditation.

National Association of Testing Authorities (NATA)

NATA is a technical accreditation provider to laboratories, and is formally recognised by the Federal Government as the national authority for accreditation of laboratories conducting tests and measurements in all technical fields. All diagnostic laboratories and pathology services will be rigorously assessed and accredited by the National Association of Testing Authorities (NATA), the independent authority for technical best practice and a requirement for Medicare Pathology billing (see Table 1). Currently, semen analysis and related testing (like sperm antibodies etc) are covered by Medicare but sperm cryostorage is not specified in the NATA accreditation and as such is not covered by Medicare.
<table>
<thead>
<tr>
<th>Table 1: Accreditation</th>
<th>Andrology</th>
<th>Accreditating Body</th>
<th>Standard</th>
<th>On-going accreditation</th>
</tr>
</thead>
<tbody>
<tr>
<td>Diagnostic Testing</td>
<td>Andrology</td>
<td>NATA</td>
<td>ISO 15189</td>
<td>Audited over a 4 year</td>
</tr>
<tr>
<td></td>
<td>(for example semen analyses and anti-sperm antibody testing)</td>
<td>Field of Testing – Medical Testing (Medicare rebates for eligible tests are only available to facilities holding NATA accreditation)</td>
<td></td>
<td>cycle involving online assessment activities, at least one Surveillance visit by a NATA lead assessor and at least one Reassessment visit involving an on-site visit by NATA lead assessor and technical assessor</td>
</tr>
<tr>
<td>Clinical Preparations</td>
<td>Andrology</td>
<td>RTAC</td>
<td>Code of Practice for Assisted Reproductive Technology Units</td>
<td>Audited by JAS-ANZ accredited certification bodies over a 3 year cycle, with minimum of yearly surveillance visits for auditing of Critical Criteria (including identification/traceability processes), whilst Good Practice Criteria auditing occurs at least once per 3 year cycle</td>
</tr>
<tr>
<td>(for example preparation of IUI samples and freezing of semen for future use)</td>
<td>(When laboratory is part of an Assisted Reproductive Technology facility)</td>
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Recommendation to PASC
Following national consultation with patients, parents, partners and health care professionals (doctors, nurses, psychologists, and counselors) and fertility laboratory staff, we are seeking three new oncofertility Medicare item numbers.

The new oncofertility Medicare item numbers will allow the development of evidence-based internationally recognised oncofertility practice in Australia as well as allowing equitable access to all patients requiring fertility preservation.

Following the creation of oncofertility Medicare item numbers, insurance agencies supported by the Federal Government, will be encouraged to incorporate oncofertility care into insurance item numbers. This will mean that patients receiving gonadotoxic treatment with medical insurance will have policies, which may cover some or all of the costs of oncofertility care.

New MBS item numbers
1. Processing and cryopreservation of ovarian tissue for fertility preservation treatment for female patients.

Population
1. Male and female patients with any cancer irrespective of stage, who will receive or have received gonadotoxic treatment in three categories paediatric, adolescent young adult and adult populations; and
2. Male and female patients with non-malignant disease who will receive or have received gonadotoxic treatment in three categories paediatric, adolescent young adult and adult populations.

Frequency of procedure
Maximum of one procedure (which may include semen being collected more than once) prior to and/or after receiving gonadotoxic treatment. Some patients may need to have fertility preservation before and after cancer treatment to ensure an adequate collection.

Restriction
Female and male patients who have fertility preservation for nonmedical indication or infertility treatment and who have not received gonadotoxic treatment.
Clinical Indications for testing in patients who will or have received gonadotoxic treatment

Newly diagnosed or relapsed patients receiving gonadotoxic treatment should have an assessment of gonadotoxic risk and need for fertility preservation prior to the start of treatment. A detailed history is required by either a physician, cancer or fertility doctor to determine the reproductive risk of the patient.

- For pre-pubertal female paediatric patients see algorithm 1a: algorithm for fertility preservation in new and relapsed paediatric female patients prior to receiving gonadotoxic treatment and 1b - algorithm for the assessment of female paediatric patient’s reproductive potential following gonadotoxic treatment
- For Adolescent Young Adult Patients (AYA) female patients (15 to 25 year old) see Algorithm 2a; algorithm for fertility preservation in new and relapsed adolescent and young adult (AYA) female patients prior to receiving gonadotoxic treatment and 2b: algorithm for the assessment of reproductive potential for adolescent young adult (AYA) female patient’s following gonadotoxic treatment
- For adult female patients see Algorithm 3a: Algorithm for fertility preservation in new and relapsed paediatric female adult patients prior to receiving gonadotoxic treatment and 3b: Algorithm for the assessment of female adult patient’s reproductive potential following gonadotoxic treatment
- For pre-pubertal male paediatric patients see 4a: Algorithm for fertility preservation in new and relapsed paediatric male patients prior to receiving gonadotoxic treatment 4b: Algorithm for the assessment of male paediatric patient’s reproductive potential following gonadotoxic treatment
- For adolescent and young adult (AYA) and adult male patients see 5a: Algorithm for fertility preservation in new and relapsed adolescent and young adult (AYA) and adult male patients prior to receiving gonadotoxic treatment and 5b: Algorithm for the assessment of male adolescent young adult and adult patient’s reproductive potential following gonadotoxic treatment.
Health Outcomes

**Fertility preservation and reproductive health**
This is the key patient-relevant outcome, for which ‘pregnancy’ is one outcome measure. AMH is also a surrogate measure for fertility following treatment with gonadotoxic therapy. Other outcomes may include the age of mother at conception (with different associated risk profiles), willingness to attempt conception.

**Miscarriage**
Treatment with gonadotoxic therapy may result in a patient becoming sub-fertile or infertile and this may have an effect on the opportunity for a woman to have a future biological pregnancy; and the late effects of gonadotoxic treatment may: (i) have a direct effect on a female patient’s egg quality, (ii) ability to hold a pregnancy; (iii) have an effect on the position of implantation of the embryo and therefore increase the chance of malposition/malpresentation, miscarriage if a female falls pregnant naturally.

**Pregnancy**
Literature supports that patients who have received treatment with gonadotoxic treatment, who have conceived naturally, tend to experience more clinical complications during their pregnancy placing them at greater risk for pregnancy and birth complications compared with women who have undergone treatment with IVF.[200, 201] Women who undergo fertility preservation prior to starting treatment with gonadotoxic therapy and then undergo assisted reproductive technologies have better pregnancy outcomes and experience less complications throughout their pregnancy compared with women who have assisted reproductive pregnancy using gonadal tissue or ametes collected following cancer treatment.

**Premature and still birth**
Babies of survivors who have natural pregnancies are reported to experience significantly elevated risks of preterm delivery, low birth weight and neonatal morbidities (including admission to a special care unit) compared to those born to women who have had undergone ART and are managed cautiously throughout the duration of pregnancy through to delivery.[202-205]

**Increased Quality of life**
The ability to have a biological family following treatment with gonadotoxic therapy, as a result of storing gametes and gonadal tissue prior to treatment has immeasurable benefits to both a patient and their partner. Patients who are given the opportunity to store their own gametes and gonadal tissue experience less psychological distress on completion of treatment compared with patients who were not given the opportunity to undergo fertility preservation.

**Improved relationships and family life**
Loss of reproductive potential after receiving gonadotoxic treatment can negatively impact relationships and family quality of life (QOL) in survivors.[161, 206] Recent studies have indicated that the potential iatrogenic loss of fertility, the loss of a potential child, has a profound impact on recipients of gonadotoxic treatment and, at times, may be more stressful than the diagnosis itself.[207] The literature reports that patients who are given the opportunity to store their own gametes and gonadal tissue experience less
psychological distress and therefore do not have the pressures of potentially becoming childless on completion of treatment as compared with patients who are not given the opportunity to undergo fertility preservation and may be rendered childless as a consequence of treatment. This can place added stresses on a relationship where the survivor would like to have a child but is unable to do so because of the detrimental effects of treatment received.
## Summary of costs

<table>
<thead>
<tr>
<th>MBS Item Number</th>
<th>Description</th>
<th>Cost</th>
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| **MBS ONC1**   | Processing and cryopreservation of ovarian tissue for fertility preservation treatment for female patients.  

**Preparation of the cortical ovarian tissue**  
The ovarian tissue is prepared prior to freezing by dissecting apart the surface (cortical) tissue containing the follicles and the inner part mainly circulation and support tissue. The surface tissue is subsequently dissected into 1mm thick slices to facilitated movement of the cryoprotectants (anti-freeze solutions). This is a manual procedure.  

**Freezing of the cortical ovarian tissue**  
The slices of ovarian cortical tissue are exposed to cryoprotectants to remove water from the cells and placed in vials in an automated freezing machine which gradually reduces temperature at a controlled rate over time to -150 °C. The vials are then stored in a large tank containing liquid nitrogen at a temperature below -150 °C. | Fee proposal:  
Cost $684 partial ovarian tissue cryopreservation.  
$1250 whole ovary ovarian tissue cryopreservation  

**Benefit:**  
Nil |
| **MBS ONC2**   | Processing and cryopreservation of semen for fertility preservation treatment. | Fee proposal:  
$ 495  

**Benefit:** |
| **MBS ON3**    | Processing and cryopreservation of testicular tissue for fertility preservation treatment. | Fee proposal:  
$ 675 |
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<th>Population</th>
<th>Intervention</th>
<th>Comparator</th>
<th>Outcomes</th>
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<tr>
<td>Female patients receiving gonadotoxic</td>
<td>MBS ONC1</td>
<td>• Infertility post-gonadotoxic treatment</td>
<td>Health Outcomes</td>
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<tr>
<td>treatment</td>
<td>Processing and cryopreservation of ovarian tissue for fertility preservation treatment for female patients.</td>
<td>• Natural pregnancy following treatment with gonadotoxic treatment</td>
<td>• Fertility preserved</td>
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<td>• Improved relationship and family life</td>
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<tr>
<td>Male patients receiving gonadotoxic</td>
<td>MBS ONC2</td>
<td>• Infertility post-gonadotoxic treatment</td>
<td>Cost-effectiveness</td>
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<td>• Cost</td>
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**Proposed structure of economic evaluation**

A full economic evaluation will be submitted following approval of this stage.
Consultation
The application for the Protocol Advisory Sub Committee Report on Oncofertility Item Numbers has had widespread consultation and support from consumers representing patients, parents and partners, advocacy groups and a range of health care providers as detailed in the list provided below:

**FUTuRE Fertility chief investigators and lead investigators**
NSW Dr Antoinette Anazodo (CI)
NSW Ms Brigitte Gerstl (AI)
NSW Professor William Ledger (CI)
NSW Professor Elizabeth Sullivan (CI)
NSW Professor Michael Chapman (CI)
NSW Associate Professor Claire Wakefield (CI)
NSW Professor Richard Cohn (CI)
NSW Dr Rebecca Deans (CI)
NSW Professor Rosalie Viney (AI)
VIC Professor Kate Stern (CI)
VIC Professor Rob Mclachlan (CI)
VIC Dr Yasmin Jayasinghe (CI)
VIC Dr Lisa Orme (CI)
VIC Ms Franca Agresta (AI)
QLD Dr Wayne Nicholls
QLD Associate Professor Anusch Yasdani
QLD Dr Ben Kroon
WA Dr Marianne Phillips
SA Professor Bogda Koczwara
SA Dr Michael Osborne
SA Dr Fiona Young
TAS Dr Rosemary Harrop

**Australasian Oncofertility Consumer Group**
NSW Ms Heather Minnich- patient representative
NSW Mr Marcus Ehrlich – patient representative
NSW Ms Rikki Hickey – partner representative
NSW Ms Jo Pedgrift – support person to consumer
QLD Dr Alex Powell - patient representative
SA Mr Mark Haseloff – patient representative
VIC Mrs Sophia HO – parent representative
WA Miss Bronwyn Kilby - patient representative

**Fertility Society of Australia Medical Fertility Preservation Group**

**Access Australia’s National Infertility Network Ltd**
Dr Sandra Dill Managing Director Access Australia’s National Infertility Network
CanTeen Youth Advisory Group
Miss Xenia Alexander, co-Chair
Mark Haseloff, co-Chair
Mr Keifer King
Mr Byron Walker
Mr Jarrod Eggins,
Miss Jenna Moloney
Miss Bronwyn Kilby
Mr Thomas Binns
Miss Jasmine Gailer
Miss Elodie Nadon
Mr Nikhil Autar

CanTeen Leadership Group
Dr Antoinette Anazodo
Dr Hera Dimitriadis
Dr Lisa Orme
Ms Kate Thompson
Dr Michael Osborn
Mr Allan Hayward
Dr Rachel Hughes
Dr Po Inglis
Ms Roslyn Henney

Youth Cancer Services Strategic Advisory Group

Andrology
Professor David Handelsman
Mr Christopher Nicolls
Professor Robert Mclachlan

IVF Directors
Professor David Molloy, Chair

Medical Oncology Group of Australia (MOGA)
Professor Rosemary Harrop, Chair

Cancer Nurse Society of Australia (CNSA)
Ms Deborah Hoberg, Chair SA
Ms Marie Condon, Chair WA
Ms Robyn Wilson, Chair VIC
Ms Lyndal Moore, Chair NSW Hunter
Ms Meredith Cummins, Chair NSW Central
South Australian Oncofertility Group

Queensland Oncofertility Group

Victorian Fertility Preservation Taskforce
References


23. *Australian Institute of Health and Welfare (AIHW).* Australian Cancer Incidence and Mortality (ACIM) books: Canberra. AIHW.


133. Barton, S.E., et al., Female cancer survivors are low responders and have reduced success compared with other patients undergoing assisted reproductive technologies. Fertility and sterility, 2012. 97(2): p. 381-386.
145. Meirow, D., et al. Results of one center indicate that transplantation of thawed ovarian tissue is effective. Repeated IVF reveals good egg quality and high pregnancy rate. OXFORD UNIV PRESS GREAT CLARENDON ST, OXFORD OX2 6DP, ENGLAND.
147. Cao, Y.-X. and R.-C. Chian. Fertility preservation with immature and in vitro matured oocytes.


Appendix 1
Australasian Oncofertility Consortium Charter

1. All cancer clinicians should discuss the possible effects of cancer treatment on a patient’s fertility before the start of treatment, irrespective of age, diagnosis and prognosis of the patient.

2. Cancer clinicians should give patients an opportunity to discuss a patient’s future fertility by offering referral to specialists who can discuss fertility preservation strategies and the fertility and reproductive health follow-up following cancer treatment.

3. Cancer centres should have a clear referral pathway between cancer and fertility and/or andrology services to ensure that a fertility preservation consultation and appropriate treatment can be organised in a timely manner when it is deemed appropriate to do so before the onset of cancer treatment.

4. National oncofertility data should be collected to enable the development and implementation of national standardised guidelines and governance structure, which takes into consideration the age of a patient.

5. Oncofertility care should be incorporated into the training curriculum for cancer and fertility multi-disciplinary health professionals at both graduate and postgraduate levels to ensure that oncofertility care becomes standard practice in Australasia.

6. Fertility preservation strategies and storage of gonadal tissue and embryos should be affordable and equitable for all cancer patients irrespective of age, ethnicity, sexual orientation or socioeconomic factors.

7. Fertility related psychosocial support should be available to all cancer patients during and after cancer therapy, irrespective of whether they pursued fertility preservation strategies.

8. Health care professionals should give all patients reproductive health information and support. This will enable patients to initiate or maintain personal relationships following a cancer diagnosis and maintain safe sexual health practices.
Figure 1a
Algorithm for fertility preservation in new and relapsed paediatric female patients prior to receiving gonadotoxic treatment

1. Identify reproductive risk concerns (gynaecological history, family history and gynaecological cancer)

2. Gonadotoxic Risk of Treatment Assessed
   - Yes: Referral for investigation for fertility preservation
   - Expected/possible: Consideration of fertility preservation as chemotherapy or novel agent may be gonadotoxic
   - No: Fertility preservation not required

3. Pre-treatment AMH and Estradiol, ultrasound for AFC

4. Fertility preservation options discussed and ovarian cryopreservation recommended

5. Ovarian cryopreservation undertaken. Process, handling and storage of ovarian tissue undertaken

6. Monitor patient after completion of gonadotoxic treatment with post-treatment: AMH/AFC/FSH
Figure: 1b Algorithm for the assessment of female paediatric patient’s reproductive potential following gonadotoxic treatment

- Identify reproductive risk concerns (gynaecological history, family history and gynaecological cancer)
- AMH taken 12 months post gonadotoxic treatment
  - Very low <0.1pmol/l
    - Check FSH with abdominal ultrasound
    - Premature ovarian insufficiency
    - Patients with persistent premature ovarian insufficiency referral for puberty induction and HRT follow-up
    - Recovering ovarian insufficiency- regular follow-up and fertility preservation if not previously offered
  - Low
    - Check FSH and abdominal ultrasound
    - If fertility preservation was not considered - referral for consideration of fertility preservation
    - Patients under 16 years of age for consideration of ovarian tissue cryopreservation or oocyte cryopreservation
  - Normal
    - Regular follow-up if symptoms of ovarian failure or prior to family planning
    - If patient has had fertility preservation monitor ovarian function annually
    - Patients over 16 years of age for consideration of ovarian tissue cryopreservation or oocyte cryopreservation
Figure 2a: Algorithm for fertility preservation in new and relapsed adolescent and young adult (AYA) female patients prior to receiving gonadotoxic treatment
Figure 2b: Algorithm for the assessment of reproductive potential for adolescent and young adult (AYA) female patient’s following gonadotoxic treatment

1. Identify reproductive risk concerns (gynaecological history, family history and gynaecological cancer)

2. Assessment of ovarian function
   AMH 12 months post gonadotoxic treatment

   a. Very low <0.1pmol/l
      - Check FSH / ultrasound
        - Premature ovarian insufficiency
        - Persistent ovarian insufficiency refer to HRT clinic and counselling

   b. Low
      - Ultrasound Check FSH
        - Recovering ovarian insufficiency: then regular follow-up and fertility preservation if not previously offered
        - If fertility preservation was not considered - referral for consideration of fertility preservation

   c. Normal
      - Regular follow-up if symptoms of ovarian failure or prior to family planning
      - If previous fertility preservation monitor ovarian function as required
Figure 3a: Algorithm for fertility preservation in new and relapsed adult female patients prior to receiving gonadotoxic treatment

1. Identify reproductive risk concerns (gynaecological history, family history and gynaecological cancer)

2. Gonadotoxic Risk of Treatment Assessed
   - Yes
     - Referral for investigation for fertility preservation
   - Expected/possible
     - Consideration of fertility preservation as chemotherapy or novel agent may be gonadotoxic
   - No
     - Fertility preservation not required

3. Pre-treatment AMH and Estradiol, FSH
   - Ultrasound for AFC

4. Fertility preservation options discussed and oocyte or embryo cryopreservation or ovarian cryopreservation undertaken if appropriate
Figure 3b: Algorithm for the assessment of reproductive potential for adult female patient’s following gonadotoxic treatment

1. Identify reproductive risk concerns (gynaecological history, family history and gynaecological cancer)

2. Fertility preservation undertaken. Process, handling and storage of gametes and gonadal tissue undertaken

3. Cancer team to notify fertility storage facility in the event of patient death so that sample is destroyed or donated to research

4. Assessment of ovarian function
   AMH 12 months post gonadotoxic treatment

   - Very low <0.1 pmol/l
     - Check FSH/ultrasound
     - Premature ovarian insufficiency
     - Persistent ovarian insufficiency refer to HRT clinic and counselling

   - Low
     - Ultrasound Check FSH
     - Recovering ovarian insufficiency then regular follow-up and fertility preservation if not previously offered

   - Normal
     - Regular follow-up if symptoms of ovarian failure or prior to family planning
     - If fertility preservation was not considered - referral for consideration of fertility preservation
     - If previous fertility preservation monitor ovarian function as required
Figure 4a: Algorithm for fertility preservation in new and relapsed paediatric male patients prior to receiving gonadotoxic treatment

1. **Identify reproductive risk concerns (family history)**

2. **Gonadotoxic Risk of Treatment Assessed**
   - **Yes**
     - Referral for investigation for fertility preservation
   - **Expected/possible**
     - Consideration of fertility preservation as chemotherapy or novel agent may be gonadotoxic
   - **No**
     - Fertility preservation not required

3. **Pre-Treatment testosterone level if appropriate**

4. **Fertility preservation options discussed and testicular cryopreservation considered**

5. **Testicular cryopreservation undertaken, process, handling and storage of testicular tissue undertaken**

6. **Cancer team to notify fertility storage facility in the event of patient death**
Figure 4b: Algorithm for the assessment of male paediatric patient’s reproductive potential following gonadotoxic treatment

1. Identify ongoing reproductive risk concerns
2. Identify gonadotoxic risk from previous treatment
3. Assessment of testicular function

Risk Identified or Expected/Possible

Patient had testicular cryopreservation, consider semen analysis, banking and storage once post pubertal

Assess testicular function and semen morphology once post pubertal

Assessment prior to consideration of family planning

Patient did not have fertility preservation prior to starting gonadotoxic treatment

Referral for fertility preservation

Semen analysis and storage once post pubertal

No risk identified

Fertility Preservation not required

Assess testicular function and semen morphology once post pubertal and then as required

Assessment prior to consideration of family planning
Figure 5a: Algorithm for fertility preservation in new and relapsed adolescent young adult and adult male patients prior to receiving gonadotoxic treatment

1. Identify reproductive risk concerns
2. Gonadotoxic Risk of Treatment Assessed
   - Yes
   - Expected/possible
     - Referral for investigation for fertility preservation
     - Consideration of fertility preservation as chemotherapy or novel agent may be gonadotoxic
       - Sperm banking options discussed. Testicular biopsy an option for patients who are very unwell or have neurological impairment.
     - Fertility preservation undertaken. Process, handling and storage of gametes and gonadal tissue undertaken
   - No
   - Fertility preservation not required
     - Follow-up post treatment and make plans for further assessment as required

3. Cancer team to notify fertility storage facility in the event of patient death
Figure 5b: Algorithm for the assessment of male adolescent young adult and adult patient’s reproductive potential following gonadotoxic treatment

1. Identify ongoing reproductive risk concerns (partner’s gynaecological history, patient's and partner's family history)
2. Identify gonadotoxic risk from previous treatment
3. Assessment of testicular function

Follow-up testicular function – semen analysis, testosterone, FSH

Risk Identified or

Patient had adequate/optimal fertility preservation before gonadotoxic treatment. No fertility preservation required.

Assess testicular function and semen morphology in all male patients to document return of normal testicular function 12 months following gonadotoxic treatment and then annually as required

Assessment prior to consideration of family planning

Patient did not have fertility preservation prior to starting gonadotoxic treatment

Referral for fertility preservation

Semen analysis and storage as required

No risk identified

Fertility Preservation not required

Assess testicular function and semen morphology in all male patients to document return of normal testicular function 12 months following gonadotoxic treatment and then annually as required

Assessment prior to consideration of family planning